ACS Sustainable Chemistry & Engineering

Antibacterial Networks Based on Isosorbide and Linalool by Photoinitiated Process

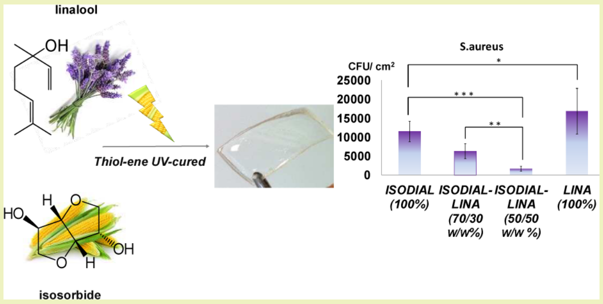
Tina Modjinou,[†] Davy-Louis Versace,[†] Samir Abbad-Andallousi,[‡] Noureddine Bousserrhine,[‡] Julien Babinot,[†] Valérie Langlois,[†] and Estelle Renard*,[†]

[†]Institut de Chimie et des Matériaux de Paris-Est, UMR 7182, CNRS-UPEC, 2-8 rue Henri Dunant, 94320 Thiais, France

[‡]Institut d'écologie et des sciences de l'environnement de Paris (IEES Paris), département SoléO (Sol et Eaux) équipe SIIM, Université Paris-Est, 61 avenue du Général de Gaulle, 94010 Créteil cedex, France

Supporting Information

ABSTRACT: In this study, we used photo-induced thiol-ene reactions to design bio-based cross-linked networks from diallyl derivative isosorbide, a crude monoterpene as linalool and a trithiol. The aim of this study is to prepare new antibacterial UV-cured networks by using a thiol-ene formulation with covalent immobilization of monoterpenes as antibacterial agents and without any organic solvent thanks to the solvent-like properties of linalool. The challenge was to demonstrate the advantage of using linalool as a chemical platform molecule and, at the same time, as an antibacterial agent. The incorporation of linear linalool to the rigid isosorbide moiety generates flexible networks. Different networks were prepared by varying the mass ratio of linalool (from 0 to 100% w/w) mixed into the coating and then characterized. Their antibacterial activities were investigated in vitro against two pathogenic bacteria strains: *Escherichia coli* and *Staphylococcus aureus*. The results have shown a promising antiadherence for *S. aureus* without any diffusion of linalool that will allow potential applications of terpenes coatings for antibacterial adhesion.



KEYWORDS: *Isosorbide, Linalool, Terpene, Thiol-ene, UV, Antibacterial network, Green chemistry*

INTRODUCTION

During the last decades, the development of bio-based molecules to replace petroleum based resources has been constantly increasing. This global effort from the scientific community is in perfect agreement with the social demands of sustainable management to reduce our reliance on fossil resources and decrease the environmental impacts of the petroleum chemistry. The development of bioplastics from renewable resources is expected to reach one million tons in 2017.[1] Among the renewable molecules of the chemical platform, terpenes represent a wide range of cheap and abundant hydrocarbons made of isoprene units (C5) that will not affect agricultural resources. Pure or crude terpenes are usually used as perfumes and fragrances. Furthermore, they are excellent solvents and diluting agents for dyes and varnishes. Even though terpenes are involved in numerous catalytic chemical processes, only a few of them (especially α-pinene, β-pinene, limonene) have been the subjects of studies related to their use as polymeric units.[2−6] Terpenes are known as components of essential oils with antibacterial and antimicrobial activities.[7−11] Such significant impacts have been observed for essential oils incorporated by blending[12−16] in a polymer matrix. The presence of microbial biofilm on polymeric surfaces represents a serious problem with substantial implications in industrial and health care processes. Several recent publications discuss the use of essential oils as additives to films or polymer matrices to produce active packaging.[12,14,17,18] However, the activity of these active polymeric materials lies in the terpenes' ability to diffuse out of the materials, which raises the issue of the sustainability of their activity over time. To overtake this drawback, we suggest to bind covalently terpenes inside a network and to evaluate the potential preservation of their specific antibacterial properties. To the best of our knowledge, there is only one study about this topic.[19] The authors report the synthesis of a polyterpineol film derived from terpinen-4-ol using radio frequency polymerization. The aim of our research is to prepare new antibacterial UV-cured networks by using a thiol-ene process with covalent immobilization of monoterpenes as antibacterial agents and without using any organic solvent. We want to demonstrate the advantage of using linalool as a chemical platform molecule and, at the same time, as an antibacterial agent.

Thiol-ene reactions involving cyclic monoterpene, limonene and multifunctional thiols[20] as promising bio-based template molecules, have been previously described without any biological investigation. Several reports on the antimicrobial

Received: January 12, 2015
Revised: March 26, 2015
Published: May 5, 2015

DOI: 10.1021/acssuschemeng.5b00018
ACS Sustainable Chem. Eng. 2015, 3, 1094−1100

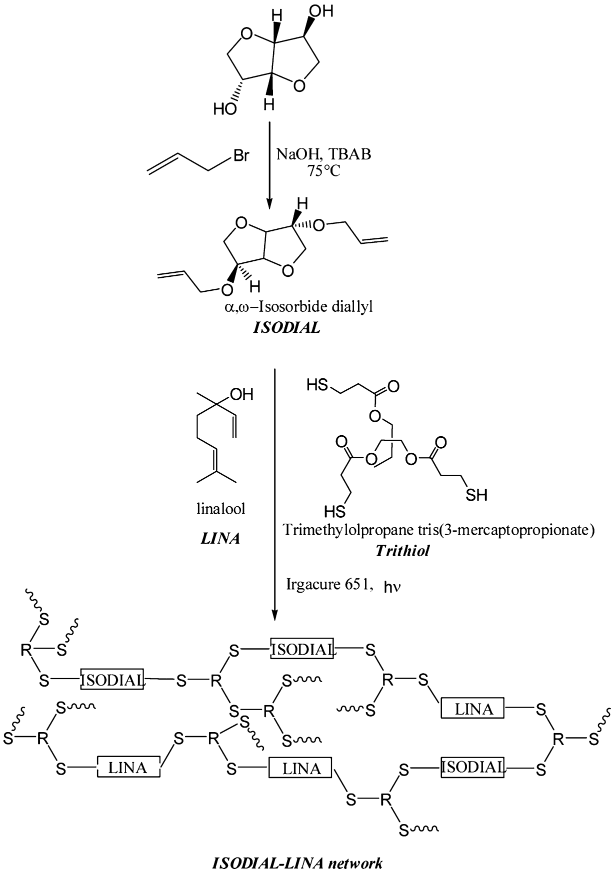
activity of some monoterpenes have shown that the number of double bonds in the structure and acyclic, monocyclic and/or bicyclic structures have no significant influence on their activity.[21,22] The relative inactivity of hydrocarbons like myrcene is associated with their low aqueous solubility while the formation of hydrogen bonds is associated with high antimicrobial activity. However, oxygenated terpenoids show distinct activities and characteristic patterns toward microorganisms.[23] Moreover, the essential oils that contain alcohol possess higher activity than those with the corresponding carbonyl compounds.[22] Based on these previous results, linalool was selected for its antibacterial properties.[9,10,23−25] Thanks to the presence of two unconjugated double bonds, it can also be used as an efficient building molecule for the preparation of networks by thiol-ene chemistry in the presence of a multifunctional thiol. Due to its linear structure and the very flexible thioether bond,[20,26−29] the association with a rigid segment is therefore essential to grant the mechanical properties of the resulting network. The bio-based rigid unit selected in the present study is isosorbide (1,4:3,6-dianhydro-D-sorbitol), which is prepared by a double dehydration of sorbitol and is available in industrial quantities. The attractive features of isosorbide are linked to its rigidity, chirality, nontoxicity and renewable origin.[30−34] Moreover, the two hydroxyl groups of isosorbide can be used directly for polycondensation or for chemical modification to prepare reactive precursors. Isosorbide can also constitute a serious alternative of diglycidyl ether bisphenol A (DGEBA) in epoxy networks, as reported. Isosorbide derivatives were used to prepare bio-based epoxy networks and the resulting structures were evaluated and compared to a conventional epoxy network based on DGEBA cured by amine curing agent, isophorone diamine.[35] In spite of the increasing interest in isosorbide, its association with terpenic molecules has not been studied yet.

To remain consistent with an eco-friendly process, UV initiated thiol-ene chemistry was chosen. This chemical route is now widely considered as a green process because it can proceed under mild conditions (room-temperature, solvent free, without metal catalysis) and fast kinetics.[36,37] This procedure is an easy and modular synthetic methodology for the preparation of networks with variable properties. The thiol-ene is an extremely versatile reaction reacting at its internal and terminal double bonds. The terminal ones are the more reactive one. These properties provided by thiol-ene UV system make it attractive for a wide range of applications: medical systems, advanced coatings or adhesives. The thiol-ene can be used with linalool but required the synthesis of isosorbide diallyl derivatives, ISODIAL, as illustrated in Scheme 1. An optimized cross-linked formulation is obtained by combining it with a trithiol-functional cross-linking agent (trimethylolpropane tris(3-mercaptopropionate)) to yield the desired network. Thermal and mechanical properties of the new bio-based networks are reported and their actions against the adhesion of both pathogenic bacteria species (*Staphylococcus aureus* and *Escherichia coli*) are evaluated and discussed thoroughly.

EXPERIMENTAL SECTION

Materials. Trimethylolpropane tris(3-mercaptopropionate) ≥95% (Trithiol), linalool 97% and sodium hydroxide Sigma Ultra minimum 98% were purchased from Aldrich. 2,2-Dimethoxy-1,2-diphenylethan-1-one (DMPA) was provided by BASF company. Allyl bromide 99% stab. with 300−1000 ppm propylene oxide was obtained from Alfa Aesar. Tetrabutylammonium bromide ≥98% (TBAB) was purchased from Fluka. Isosorbide was provided by Roquette. Dichloromethane and diethyl ether used were of analytical grade. Other chemicals for this study were analytic reagents obtained from Aldrich and used without purification.

Scheme 1. Synthesis of ISODIAL and Formation of the ISODIAL-LINA Network by Thiol-ene Reaction



Synthesis of α,ω-Isosorbide Diallyl, ISODIAL. α,ω-Isosorbide diallyl was synthesized by the Williamson reaction.[38,39] Isosorbide (3 g, 20.5×10^{-3} mol) and tetrabutylammonium bromide (300 mg, 9.3×10^{-4} mol) were dissolved in 12 mL of an aqueous NaOH solution (6 g, 1.5×10^{-1} mol, 2.10^{-3} g·L^{-1}). Allyl bromide (10 mL, 1.2×10^{-4} mol) was added to the mixture as an alkylating agent. The mixture was heated at 75 °C for 4 h 30 and then extracted with dichloromethane. The organic phase was washed with 100 mL of 1 M HCl to neutralize the solution. Afterward, several water washes were carried out to remove the salts. The organic layer was then dried under $MgSO_4$ and evaporated to give the crude product with a yield of 80−90%.

Isosorbide−Linalool Networks Preparation, ISODIAL-LINA Networks. As an example, the synthesis of a 70−30 (w/w %) α,ω-isosorbide diallyl−linalool network (1 eq SH/C═C) is described here. Linalool (103 μL, 90 mg, $5.8.10^{-4}$ mol) and DMPA (1.5 wt % of total mixture) were dissolved in α,ω-isosorbide diallyl (210 mg, 9.3×10^{-4} mol). Trithiol (398 mg, 9.9×10^{-4} mol, 1 eq SH/C═C) were added to the solution. The formulation was deposited into a silicone mold (1.8 × 4 cm) and irradiated 5 min at room temperature with a Hamamatsu Lightning LC8 (L8251) instrument. It was equipped with a mercury−xenon lamp (200 W) coupled with a flexible light guide. The end of the guide was placed 11 cm from the sample's surface. The maximum UV light intensity at the sample position was measured by radiometry (International Light Technologies ILT 393). It was 180 mW cm^{-2} in the 250−450 nm. A transparent network was obtained.

Characterization. ^{1}H NMR (400 MHz) spectra were recorded on a Bruker AV 400M in $CDCl_3$ at 25 °C. Fourier transform infrared (FTIR) spectra were recorded on a Bruker Tensor 27 spectrometer with 32 scans equipped with an attenuated total reflectance (ATR) apparatus.

Kinetics of the photoaddition reactions were followed by real-time Fourier transform infrared spectroscopy (RT-FTIR) using a Thermo-Nicolet 6700 instrument and a 80 μL of a liquid mixture composed of ISODIAL, linalool and trithiol. The photoinitiator (DMPA) was dissolved to a 3% w/w dilution. This sample was applied to a BaF_2 chip by a calibrated wire-wound applicator. The thickness of the UV-curable film was evaluated at 4 μm. The RT-FTIR analyses were carried out under laminated conditions: a polypropylene film was deposed on the top of the photosensitive layer to prevent oxygen diffusion. Samples were simultaneously exposed for a few seconds to both the UV beam, which starts the photoreaction, and the IR beam, which analyzes the extent of the photoreaction in situ. The mixture was irradiated at room temperature with the Hamamatsu lightening cure LC8 (L8251), equipped with a mercury−xenon lamp (200 W) coupled with a flexible light guide. The end of the guide was placed at a distance of 11 cm from the sample's surface. The light intensity on the surface of the sample was about 180 mW/cm^2. The thiol-ene photoaddition was monitored by the disappearance of the C═C bonds of (I) at 1647 cm^{-1} and of the SH bonds of the trithiol at 2560 cm^{-1}.

Measurements were performed on a Horiba Xplora confocal Raman microscope (Horiba Jobin Yvon) equipped with an external excitation source (638 nm He−Ne laser). The objective lenses used in the study were 50× long-working distance (Long Working Distance M Plan Semi-Apochromat, LMPLFL50x, Olympus) and 10× (M Plan Achromat, MPLN10x, Olympus), operating in air. Spectral acquisition was obtained using LabSpec software (Horiba Scientific, Edison, New Jersey). Raman spectra were recorded at a resolution of 1 cm^{-1} in the range between 4000 and 200 cm^{-1}. The Raman analyses were performed on a dark background at 20 °C. Repeated acquisitions using the highest magnification were accumulated to improve the signal-to-noise ratio. Spectra were calibrated using the 520.5 cm^{-1} line of a silicon wafer. Spectral details: objective, 50×; filter, 100%; exposition, 10; slit, 50; hole, 500. Spectra were averaged from two to five accumulations, each with an acquisition time ranging from 15 s to 5 min. The conditions of measurement were adjusted depending on the type of the matrices to obtain a good signal-to-noise ratio.

The extent of conversion was measured via FT-Raman spectroscopy from the band 2600 cm^{-1} corresponding to the SH bonds after the normalization of the spectrum to the ester carbonyl peak at 1735 cm^{-1}. The relative conversion was calculated according to the following equation:

$$\alpha = \left(\frac{R_0 - R_t}{R_0}\right) \times 100 \qquad \text{(eq 1)}$$

with

$$R_0 = \frac{I_{\mathrm{SH}}}{I_{\mathrm{C=0}}}, \quad \text{before irradiation}$$

$$R_t = \frac{I_{\mathrm{SH}}}{I_{\mathrm{C=0}}}, \quad \text{after irradiation}$$

Water contact measurements were performed with the drop shape analysis system Krûss Easy DROP contact angle measuring system apparatus controlled by the DSA software.

The thermal properties of the networks were measured by differential scanning calorimetry (DSC). The measurements were conducted on PerkinElmer Diamond DSC apparatus according to the following heating programs: heating from −60 to +170 °C at 20 °C·min^{-1}, cooling to −60 °C at 200 °C·min^{-1}, holding at 60 °C for 5 min and heating to 170 °C at 20 °C·min^{-1} to determine the glass transition temperatures.

Thermogravimetric analyses (TGA) were performed on a Setaram Setsys Evolution 16 apparatus by heating from 20 to 800 °C at 10 °C·min^{-1} under argon atmosphere.

Water Uptake Determination. Prior to the water uptake measurements, the samples were dried under vacuum. They were placed in a conditioning closed chamber over a saturated solution of NaCl to obtain a water-saturated atmosphere of 75% at room temperature. The percentage uptake of moisture into the material was measured by a coulometric method at $t = 0$ s and after 18 h. The 831 KF Coulometer was equipped with a 728 stirrer and an 860 KF Thermoprep.

Soluble Extract Determination. The samples were treated with 10 mL of diethyl ether stirred in a flask for 1 h at room temperature. The remaining residue and the film were weighted after solvent evaporation under vacuum.

Bacterial Adhesion. The antiadherence activity of the networks was evaluated using two pathogenic bacterial strains: *Staphylococcus aureus* ATCC6538 (Gram-positive) and *Escherichia coli* ATCC25922 (Gram-negative) that were grown aerobically at 37 °C overnight on LB medium (Lysogeny Broth) before the bacterial adhesion tests. LB medium was purchased from Aldrich.

The samples of ISODIAL-LINA networks were immersed for 1 h at 37 °C and stirred in the bacterial suspension of the two different strains ($OD_{600\ \mathrm{nm}}$ = 0.05). The nonadherent bacteria were then removed from the network surface by several washes with a physiological saline buffer. The networks were then submitted to vortex and ultrasounds in a minimum volume of saline buffer to unhook fasten bacteria. Finally, 100 μL of this resulting viable bacteria suspension was inoculated onto the surface of a PCA agar (Plate Count Agar) Petri dish. After 24 h of incubation at 37 °C in aerobic conditions, the antibacterial activity was measured by counting colony-forming unit (CFUs) for the different strains. Each experiment was performed in six replicates.

The software R was used to analyze the data. At least six replicates of the experiments were conducted in order to allow a significant statistical analysis. The ANalysis Of the VAriance (ANOVA) statistical test was used and significant differences ($p < 0.05$) among antibacterial properties of the networks were detected thanks to the multiple range test of Duncan.

■ RESULTS AND DISCUSSION

Preparation of ISODIAL-LINA Networks. The present work focuses on the photoinduced thiol-ene cross-linking involving different components (Scheme 1): linalool, an unsaturated monoterpene, ISODIAL, an allylic derivative of isosorbide, the trithiol and 3 wt % of DMPA. The first step consisted in the synthesis of ISODIAL by reaction of isosorbide with allyl bromide under phase-transfer catalysis conditions as described in a previous report.[40] The chemical structure of ISODIAL is confirmed by ^{1}H NMR (see the Supporting Information) with the appearance of new signals at 5.2−5.3 and 5.8−5.9 ppm characteristic of unsaturated allyl groups. Fourier transform infrared spectroscopy (FTIR) was used to confirm this. Different peaks indicate the presence of ═CH bond (3079 cm^{-1}), C═C bond (1647 cm^{-1}), C—O bond from aliphatic ether (1059 cm^{-1}) and −CH− bond (2930, 2870 cm^{-1} and 1460, 1360 cm^{-1}).

Figure 1 shows the FTIR spectra of linalool, ISODIAL and a 70/30 ISODIAL/LINA mixture before irradiation. The absorption of SH groups can be identified at 2560 cm^{-1} and the stretching band of C═C groups for both ISODIAL and linalool appears at 1647 cm^{-1}. After irradiation, the absorbance of the SH and C═C groups should decrease, thus assuming the success of the thiol-ene reaction. Figure 2 represents the kinetic profiles (disappearance of the SH bonds) of three compositions of ISODIAL (from 100 to 50%) and of linalool (from 0 to 50%), to highlight the thiol-ene process. High SH conversions are obtained after 100 s of irradiation in the case of the ISODIAL-based network whereas a slight decrease occurred when the linalool content was increased. In the samples series with 30 and 50% of linalool, the SH conversion reached 90 and

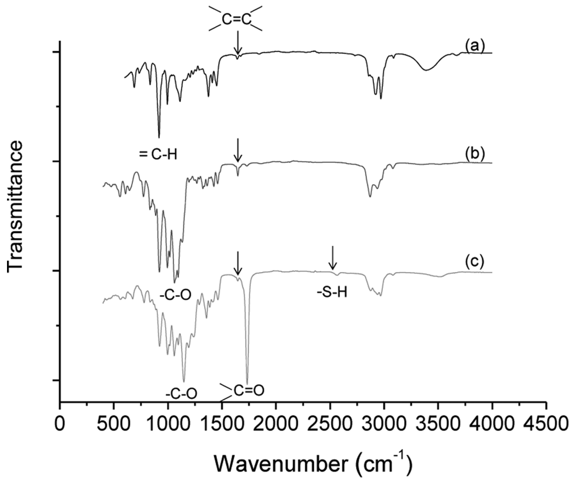


Figure 1. FTIR spectra of (a) linalool, (b) ISODIAL and (c) 70–30 ISODIAL-LINA in the presence of trithiol before irradiation.

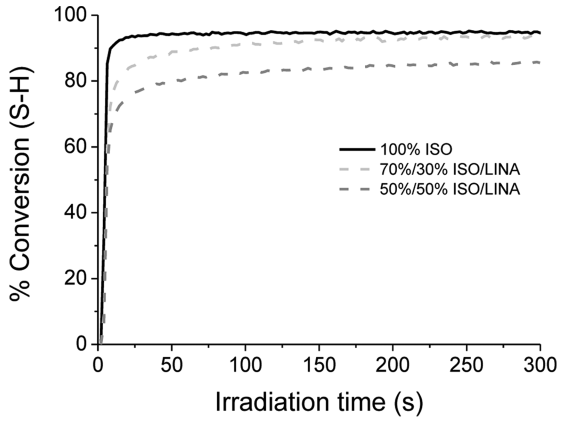


Figure 2. Real-time FTIR kinetic profiles of the formation of ISODIAL-LINA networks 100:0, 70:30 and 50:50 with DMPA in laminated conditions with Xe–Hg lamp.

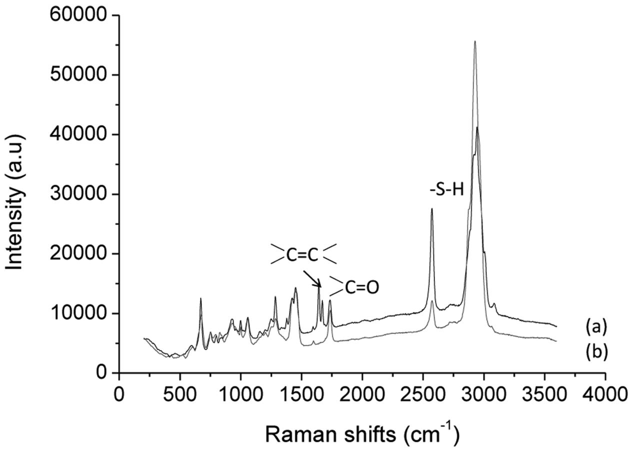


Figure 3. Raman spectra of ISODIAL-LINA 50–50 (a) before and (b) after 300 s of irradiation.

Table 1. Composition and Properties of the Networks

ISODIAL-LINA (w/w%)	100-0	70-30	50-50	0-100
Thickness (mm)	0.29 ± 0.07	0.44 ± 0.07	0.43 ± 0.09	nd
SH Conversion [a] (%)	77	78	75	73
Soluble extract [b] (%)	1	2	3	nd
				

[a] Determined by Raman. [b] Determined by gravimetry.

85%, respectively. Furthermore, it can be noticed that the thiol conversion appears slightly lower when the linalool percentage increases. This is likely due to the different reactivity of the double bonds of linalool as previously reported for limonene.[41] However, it appears that a rapid free-radical thiol-ene cross-linking occurs for all the studied compositions.

At the same time, Raman spectroscopy has been used to follow the cross-linking process of our systems. When tested, the FT-Raman spectra of the ISODIAL-LINA mixtures showed strong characteristic peaks for polarizable SH bonds and low characteristic peaks for the polar carbonyl groups (1700 cm^{-1}). It led to easier conversion measurements (Figure 3).

To assess the influence of the linalool content on the network properties, a series of photocured samples containing 0, 30, 50 and 100% of linalool were prepared. All compositions produced homogeneous viscous solutions, which were then introduced into a silicone mold and induced the formation of soft networks after UV irradiation. As illustrated in Table 1, the colorless status of crude linalool combines with its intrinsic properties as cyclic structure and chirality of isosorbide caused the transparency of the resulting isosorbide-diallylic-linalool bio-based networks. Furthermore, all the compositions with isosorbide produced transparent and soft networks, demonstrating the interest of the insertion of this rigid unit to process materials. The thickness of the samples increased from 0.29 to 0.44 mm with the ratio of linalool, which suggests a variation of the density of these networks. The 100% linalool formulation led to a very sticky network that can be removed from the silicon mold but cannot be manipulated because of its stickiness.

As reported in Table 1, the conversion of SH bonds was lower than 80% in comparison with the one measured in laminar conditions (90%). The high viscosity could explain the lack of observed final conversion. A larger amount of components decreases the mobility of reactive groups and consequently decreases the final conversion. Whatever the stoichiometry used, a total conversion was not achieved. However, the soluble fraction measured gravimetrically after extraction in appropriate solvent was lower than 5%, which indicates a high cross-linking density. These results are in agreement with the high conversion values. It can be assumed that the decreases in mobility at high final conversion restrict the accessibility of the reactive groups.

Characterization of ISODIAL-LINA Networks. Surface properties of the networks were examined by water contact angle measurements (Table 2 and Figure 4). As it was expected, the water contact angle increased with linalool addition due to the presence of apolar linalool. Furthermore, the water sensitivity of networks from hygroscopic isosorbide was investigated by measuring the amount of water taken up by conventional Karl Fisher titration. After 18 h of exposure to an atmosphere with about 75% of relative humidity, the water

Table 2. Characterization of ISODIAL-LINA Networks

ISODIAL-LINA (w/w %)	T_g[a] (°C)	$T_{5\%}$[b] (°C)	γ[c] (deg)	E[d] (Mpa)	σ_r[d] (Mpa)	ε_r[d] (%)	water uptake[e] (%)
100/0	−2	315	54	5.4 ± 0.2	2.0 ± 0.4	43 ± 8	0.5
70/30	−8	292	94	3.0 ± 0.4	1.4 ± 0.2	52 ± 7	0.7
50/50	−12	277	94	1.7 ± 0.1	0.8 ± 0.1	59 ± 7	0.7
0/100	−25	274	98	nd	nd	nd	nd

[a] Determined by DSC. [b] $T_{5\%}$: temperature of 5% weight loss of the networks determined by TGA. [c] Determined by water contact angle measurements. [d] Determined by tensile measurements. [e] Determined by coulometry after 18 h of exposure.

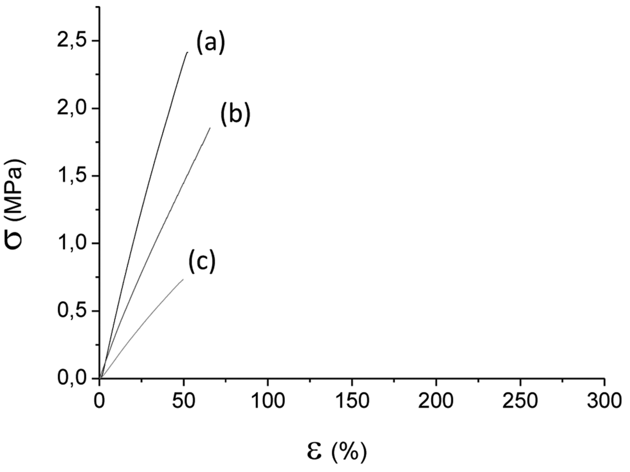


Figure 4. Tensile tests of (a) ISODIAL-LINA 100−0, (b) ISODIAL-LINA 70−30 and (c) ISODIAL-LINA 50−50.

uptakes remained lower than 5%, which indicates that samples were not sensitive to humidity. This information is important concerning the conservation of mechanical integrity when exposed to aqueous medium and for potential degradation reactions.

The thermal and mechanical properties investigated are reported in Table 2. The glass transition temperature is below 0 °C, indicating that the structure is very flexible due to the formation of flexible thioether linkages as reported previously.[20,42,43] The T_g values decrease from −2 to −25 °C, which can be attributed to the chain flexibility afforded by the higher linalool fraction. Thermal degradation was calculated at temperature corresponding to 5% mass loss. The thermal degradation temperatures of ISODIAL-LINA networks were between those of linalool and ISODIAL networks. The presence of isosorbide units increases the thermal stability.

Results obtained from stress−strain curves are presented in Figure 4 and listed in Table 2. The mechanical properties of the ISODIAL-LINA networks were tested to determine Young's modulus (E), the strain (ε) and tensile stress (σ_r). An increase in stiffness of the materials is observed thanks to the increase in cross-link density and rigid isosorbide amount, which provides manipulable networks. In addition, similar trends were observed in the tensile properties of the materials, reflecting the interplay between the linear soft linalool units and the cross-link density. The physical properties should be attuned with presynthesis of a polyfunctional resins combined with thiol cross-linkers and linalool as reported recently with limonene.[20] However, the major focus of our present report is to demonstrate the potential benefit of the incorporation of linalool in the network formulation to inhibit the bacterial adhesion.

Bacterial Adherence. The adherence ability of each studied bacteria on active networks was measured after incubation within a bacterial suspension for 1 h. The number of viable bacteria adhering to the surface (CFU/cm^2) was calculated by performing an ANOVA and a StatAdvisor post-tests (Figure 5). First, it is worth noting that the results obtained with 100% linalool are delicate to analyze in comparison with the other composition because of the sticky feature of the surface. The bacterial adhesion measurements were enhanced by the fact that bacteria remained adhered. However, the influence of the chemical composition networks on bacterial adherence was different for both bacterial strains. *E. coli* was not affected by the presence of linalool because the percentage of adherence of *E. coli* was not significantly different when we compared the adherence assay performed on 100% Iso, 70/30 ISODIAL-LINA and 50/50 ISODIAL-LINA ($p >$ 0.05). In the case of 100% linalool networks, the adherence of *E. coli* is significantly higher than these other ones ($p < 0.05$). These results could be explained in part by the sticky feature of linalool promoting the adhesion of cells on its surface. On the other hand, significant differences ($p < 0.05$) were observed with *S. aureus*. The adhesion of *S. aureus* depends on the quantity of linalool: a reduction of 45 to 85% in the bacterial adhesion was observed with proportions of respectively 30% and 50% of linalool. The CFU/cm^2 values in the case of *E. coli* were systematically lower than those obtained with *S. aureus*. It indicates that the systems based on isosorbide alone or associated with linalool present intrinsic antifouling properties. The structure of the cell wall of *E. coli* is much more complicated than the one of the Gram-positive species. There is an outer membrane mainly made of phospholipids, proteins, and lipopolysaccharides (LPS).[44,45] One of the key functions of this outer membrane is to protect the bacteria from attacks by foreign substances such as antibacterial agents. In addition, the hydrophobicity of an antibacterial polymer agent containing long alkyl chains makes it difficult to eliminate *E. coli* because of the lower interactions between the bacteria and its outer

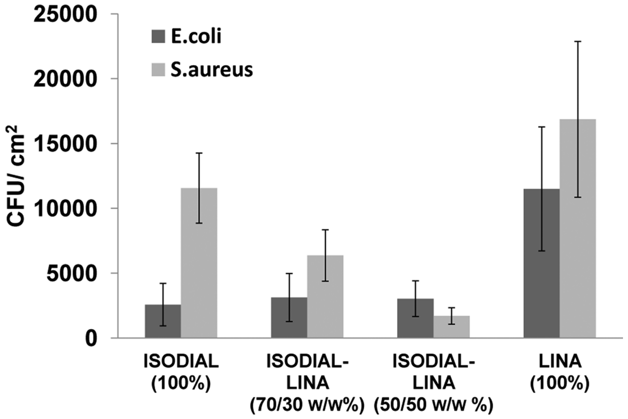


Figure 5. Antibacterial tests of ISODIAL-LINA networks.

membrane. It can also be assumed that the hydrophobicity of the surface is the main cause of the weaker attraction between *E. coli* and the surface of the network. In the case of *S. aureus*, which exhibits a preference for attachment on hydrophobic surfaces,[19,46] the antiadherence activity can be induced by the linalool. Our results agree with previous reports which incorporate essential oils into quince mucilage films.[17] The authors showed that their active packaging were more effective against Gram-positive bacteria than Gram-negative ones. Nevertheless, most of the results reported in the literature vary considerably according to the nature of the essential oils and the bacterial strains. Indeed, some essential oils containing linalool have exhibited antibacterial activity against both Gram-negative bacteria and Gram-positive bacteria.[9,47] The mechanism of action of essential oils has not been clearly identified but it seems to be related to the hydrophobic nature of the terpenes.[15] Furthermore, the numerous components of the oils could have synergic or additive effects on the bacterial activity.[47,48] Our study presents the advantage of highlighting that the linalool cross-linking coatings represent a promising route to be more resistant to the colonization of microorganisms.

■ CONCLUSION

In conclusion, the efficiency and abilities of thiol-ene reaction were used to prepare new bio-based antibacterial materials. This very fast and easily performed reaction occurs under room temperature conditions and is solvent free, which is in line with green chemistry. New bio-based materials from sustainable isosorbide moieties and natural terpene were successfully engineered according to thiol-ene photoinitiated process. ISODIAL-LINA networks exhibit attractive thermal properties and furthermore the highest inhibition observed against *S. aureus*. Moreover, these new bio-based materials are not affected by water penetration under high moisture conditions. Using natural terpenes as cross-linking units represents a promising route for the preparation of new efficient bioactive materials. It will allow further investigations with varying terpenes units and rigid comonomers in order to develop antifouling applications. However, in the future, it may be appropriate to associate different terpenes to the combination to obtain synergic effects on various bacteria strains.

■ ASSOCIATED CONTENT

Supporting Information

The ^{1}H NMR spectrum of ISODIAL and the FTIR spectra of isosorbide and ISODIAL are obtainable. The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acssuschemeng.5b00018.

■ AUTHOR INFORMATION

Corresponding Author

*E. Renard. Phone: 33 (0)1 49 78 12 17. E-mail: renard@icmpe.cnrs.fr.

Notes

The authors declare no competing financial interest.

■ ACKNOWLEDGMENTS

We thank CNRS and UPEC for financial support. The authors thank the French Ministry of Research for providing Tina Modjinou with a grant.

■ REFERENCES

(1) http://www.europabio.org.

(2) Pa, W.; Chu, F.; Tang, C. Progress in renewable polymers from natural terpenes, terpenoids, and rosin. *Macromol. Rapid Commun.* **2013**, *1* (34), 8−37.

(3) Holmberg, A. L.; Reno, K. H.; Wool, R. P.; Epps, T. H. Biobased building blocks for the rational design of renewable block polymers. *Soft Matter* **2014**, *10*, 7405−7424.

(4) Barros, M. T.; Petrova, K. T.; Ramos, A. M. Potentially biodegradable polymers based on α- or β-pinene and sugar derivatives or styrene, obtained under normal conditions and on microwave irradiation. *Eur. J. Org. Chem.* **2007**, *2007* (8), 1357−1363.

(5) Firdaus, M.; Meier, M. A. R.; Biermann, U.; Metzger, J. O. Renewable co-polymers derived from castor oil and limonene. *Eur. J. Lipid. Sci. Technol.* **2014**, *116* (1), 31−36.

(6) Singh, A.; Kamal, M. Synthesis and characterization of polylimonene: Polymer of an optically active terpene. *J. Appl. Polym. Sci.* **2012**, *125* (2), 1456−1459.

(7) Nostro, A.; Ribaltone, S.; D'Arrigo, M.; Botta, L.; Filucamo, A.; Antrazzoli, M.; Bisignano, G. Development and characterization of essential oil component-based polymer films: A potential approach to reduce bacterial biofilm. *Appl. Microbiol. Biotechnol.* **2013**, *97* (21), 9515−9523.

(8) Lang, G.; Buchbauer, G. A review on recent research results (2008−2010) on essential oils as antimicrobials and antifungals. A review. *Flavour Fragrance J.* **2012**, *27*, 13−39.

(9) Iacobellis, N. S.; Lo Cantore, P.; Capasso, F.; Senatore, F. Antibacterial activity of *Cuminum cyminum* L. and *Carum carvi* L. essential oils. *J. Agric. Food Chem.* **2005**, *53*, 57−61.

(10) Kubo, I.; Fujita, K.-I.; Kubo, A.; Nihei, K.-I.; Ogura, T. Antibacterial activity of Coriander volatile compounds against *Salmonella choleraesuis*. *J. Agric. Food Chem.* **2004**, *52*, 3329−3332.

(11) Shah, A. A.; Wang, C.; Chung, Y.-R.; Kim, J.-Y.; Choi, E.-S.; Kim, S.-W. Enhancement of geraniol resistance of *Escherichia coli* by MarA over expression. *J. Biosci. Bioeng.* **2013**, *115* (3), 253−258.

(12) Arrieta, M. P.; Lopez, J.; Hernandez, A.; Rayon, E. Ternary PLA-PHB-limonene blends intended for biodegradable food packaging applications. *Eur. Polym. J.* **2014**, *50*, 255−270.

(13) Mourtzinos, I.; Kalogeropoulos, N.; Papadakis, S. E.; Konstantinou, K.; Karathanos, V. T. Encapsulation of nutraceutical monoterpenes in β-cyclodextrin and modified starch. *J. Food Sci.* **2008**, *73* (1), 89−94.

(14) Robertson, M. L.; Paxton, J. M.; Hillmyer, M. A. Tough blends of polylactide and castor oil. *ACS Appl. Mater. Interfaces* **2011**, *3*, 3402−3410.

(15) Sanchez-Gonzalez, L.; Chafer, M.; Hernandez, M.; Chiralt, A.; Gonzalez-Matinez, C. Antimicrobial activity of polysaccharide films contaning essential oils. *Food Control* **2011**, *22*, 1302−1310.

(16) Tackenberg, M. W.; Marmann, A.; Thommes, M.; Schuchmann, H. P.; Kleinebudde, P. Orange terpenes, carvacrol and α-tocopherol encapsulated in maltodextrin and sucrose matrices via batch mixing. *J. Food Eng.* **2014**, *135*, 44−52.

(17) Jouki, M.; Yazdi, F. T.; Mortazavi, S. A.; Koocheki, A. Quince seed mucilage films incorporated with oregano essential oil: Physical, thermal, barrier, antioxydant and antibacterial properties. *Food Hydrocolloids* **2014**, *36*, 9−19.

(18) Bolton, J. M.; Hillmyer, M. A.; Hoye, T. R. Sustainable thermoplastic elastomers from terpene-derived monomers. *ACS Macro Lett.* **2014**, *3*, 717−720.

(19) Bazaka, K.; Jacob, M. V.; Truong, V. K.; Crawford, R. J.; Ivanova, E. P. The effect of polyterpenol thin film surfaces on bacterial viability and adhesion. *Polymers* **2011**, *3* (1), 388−404.

(20) Claudino, M.; Mathevet, J.-M.; Jonsson, M.; Johansson, M. Bringing D-limonene to the scene of bio-based thermoset coating via free-radical thiol-ene chemistry: Macromonomer synthesis, UV-curing and thermo-mechanical characterization. *Polym. Chem.* **2014**, *5*, 3245−3260.

(21) Zygadlo, J. A.; Maestri, D. M.; Lamarque, A. L.; Guzmán, C. A.; Velasco Negueruela, A.; Pérez-Alonso, M. J.; García-Vallejos, M. C.;

Grosso, N. R. Essential oil variability of *Minthostachys verticillata*. *Biochem. Syst. Ecol.* **1996**, *24* (4), 319−323.

(22) Zygadlo, J. A.; Juliani, H. R. Bioactivity of essential oil components. *Curr. Top. Phytochem.* **2000**, *3*, 204−214.

(23) Habibi, Z.; Yousefi, M.; Mohammadi, M.; Eftekhar, F.; Biniyaz, T.; Rustaiyan, A. Chemical composition and antibacterial activity of the volatile oils from *Artemisia turcomanica*. *Chem. Nat. Compd.* **2010**, *46* (5), 819−821.

(24) Lo Cantore, P.; Iacobellis, N. S.; De Marco, A.; Capasso, F.; Senatore, F. Antibacterial activity of *Coriandrum sativum* L. and *Foeniculum vulgare* Miller Var. *vulgare* (Miller) essential oils. *J. Agric. Food Chem.* **2004**, *52*, 7862−7866.

(25) Park, S.-N.; Kyong Lim, Y.; Oliviera Freire, M.; Cho, E.; Jin, D.; Kook, J.-K. Antimicrobial effect of linalool and α-terpineol against periodontopathic and cariogenic bacteria. *Anaerobe* **2012**, *18*, 369−372.

(26) Lecamp, L.; Houllier, F.; Youssef, B.; Bunel, C. Photoinitiated cross-linking of a thiol−methacrylate system. *Polymer* **2001**, *42* (7), 2727−2736.

(27) Nilsson, C. *Genetic Studies of Psoriasis and Psoriatic Arthritis*; Sahlgrenska Academy at University of Gothenburg: Gothenburg, Sweden, 2008.

(28) Nilsson, C.; Malmström, E.; Johansson, M.; Trey, S. Dendrimers in thiol-ene crosslinked networks and the effect of subsequent generations on thermoset properties. *J. Polym. Sci., Part A: Polym. Chem.* **2009**, *47* (2), 589−601.

(29) Nilsson, C.; Malmström, E.; Johansson, M.; Trey, S. M. Methacrylated dendrimers in thiol-methacrylate networks and the effect of conversion on the thermoset properties. *J. Polym. Sci., Part A: Polym. Chem.* **2009**, *47* (21), 5815−5826.

(30) Stoss, P.;Hemmer, R. *Advances in Carbohydrate Chemistry and Biochemistry*; Academic Press Limited: London, 1991; pp 93−173.

(31) Thiem, J.; Lûders, H. Synthesis of polyterephthalates derived from diahydroxitols. *Polym. Bull.* **1984**, *11* (3), 365−369.

(32) Chatti, S.; Weidner, S.; Fildier, A. Copolyesters of isosorbide, succinic acid and isophtalic acid: biodegradable hight Tg engineering plastics. *J. Polym. Sci., Part A: Polym. Chem.* **2013**, *51* (11), 2464−2471.

(33) Kricheldorf, H. R. "Sugar diols" as building blocks of polycondensates. *J. Macromol. Sci. Rev. Macromol. Chem. Phys.* **1997**, *37*, 599−631.

(34) Kricheldorf, H. R.; Behnken, G.; Sell, M. Influence of isosorbide on glass-transition temperature and crystallinity of poly(butylene terephtalate). *J. Macromol. Sci., Part A: Pure Appl. Chem.* **2007**, *44* (7), 679−684.

(35) Chrysanthos, M.; Galy, J.; Pascault, J.-P. Preparation and properties of bio-based epoxy networks derived from isosorbide diglycidyl ether. *Polymer* **2011**, *52* (16), 3611−3620.

(36) Lowe, A. B. Thiol-ene "click" reactions and recent applications in polymer and materials synthesis. *Polym. Chem.* **2010**, *48* (1), 17−36.

(37) Jing, X.; Yue, J.; Li, X.; Mo, G.; Wang, R.; Huang, Y. Modular functionalization of amphiphilic block copolymers via radical-mediated thiol-ene reaction. *Macromolecules* **2010**, *43* (23), 9645−9654.

(38) Feng, X.; East, A. J.; Hammond, W. B.; Zhang, Y.; Jaffe, M. Overview of advances in sugar-based polymers. *Polym. Adv. Technol.* **2011**, *22*, 139−150.

(39) Lorenzini, C.; Versace, D. L.; Gaillet, C.; Lorthioir, C.; Boileau, S.; Renard, E.; Langlois, V. Hybrid networks derived from isosorbide by means of thiol-ene photoaddition and sol-gel chemistry. *Polymer* **2014**, *55*, 4432−4440.

(40) Lorenzini, C.; Renard, E.; Bensemhoun, J.; Babinot, J.; Versace, D.-L.; Langlois, V. High glass transition temperature bio-based copolyesters from poly(3-hydroxybutyrate-co-3-hydroxyvalerate) and isosorbide. *React. Funct. Polym.* **2013**, 73 (12), 1656−1661.

(41) Meier, M. A. R.; Firdaus, M.; Montero de Espinosa, L. Terpene-based renewable monomers and polymers via thiol-ene additions. *Macromolecules* **2011**, *44* (18), 7253−7262.

(42) Carlsson, I.; Harden, A.; Lundmark, S.; Manea, A.; Rehnberg, N.;Svensson, L. Allyl ethers in the thiol-ene reaction. In *Photoinitiated Polymerization*; Belfield, K. D., Crivello, J. V., Eds.; ACS Symposium Series; American Chemical Society: Washington, DC, 2003; Vol. *847*, pp 65−75.

(43) Sangermano, M.; Colucci, G.; Fragale, M.; Rizza, G. Hybrid organic−inorganic coatings based on thiol-ene systems. *React. Funct. Polym.* **2009**, *69* (9), 719−723.

(44) Franklin, T. J.; Snow, G. A. *Biochemistry of Antimicrobial Action*; Chapman & Hall LTD: London, 1981.

(45) Sussman, M. *Escherichia Coli: Mechanisms of Virulence*; University of Cambridge Press: Cambridge, 1997.

(46) James, N. R.; Jayakrishnan, A. Surface thiocyanation of plasticized poly(vinyl chloride) and its effect on bacterial adhesion. *Biomaterials* **2003**, *24* (13), 2205−2212.

(47) Koutsoudaki, C.; Krsek, M.; Rodger, A. Chemical composition and antibacterial activity of the essential oil and the gum of *Pistacia lentiscus Var. chia*. *J. Agric. Food Chem.* **2005**, *53*, 7681−7685.

(48) Aiemsaard, J.; Aiumlamai, S.; Aromdee, C.; Taweechaisupapong, S.; Khunkitti, W. The effect of lemongrass oil and its major components on clinical isolate mastitis pathogens and their mechanisms of action on *Staphylococcus aureus* DMST 4745. *Res. Vet. Sci.* **2011**, *91*, 31−37.